

Reproduction of a Protocell by Replication of Minority Molecule in Catalytic Reaction Network

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Abstract

For understanding the origin of life, it is essential to explain the development of a compartmentalized structure, which undergoes growth and division, from a set of chemical reactions. In this study, a hypercycle with two chemicals that mutually catalyze each other is considered in order to show that the reproduction of a protocell with a growth-division process naturally occurs when the replication speed of one chemical is considerably slower than that of the other chemical. It is observed that the protocell divides after a minority molecule is replicated at a slow synthesis rate, and thus, a synchrony between the reproduction of a cell and molecule replication is achieved. The robustness of such protocells against the invasion of parasitic molecules is also demonstrated.

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Filling a gap between just a set of catalytic reactions and a reproducing cell is a difficult problem; however, it is essential to fully understand this relationship to unveil the origin of life, to establish a theory for a living state, and to experimentally synthesize protocells[1]. Several theoretical attempts to understand the relationship have been made by using catalytic networks[2–5]. For reproduction of cells, it is necessary to replicate functional polymers. At a primitive stage of life, however, errors in the sequence of polymers are inevitable during their replications, which prevent the transfer of information through replications. This was pointed out by Eigen and Schuster as an error catastrophe[4]. To resolve this error-catastrophe problem, they[4] proposed a hypercycle in which different molecular species mutually catalyzed the replication of each other. This hypercycle, however, introduced another problem, i.e., it led to the emergence of parasitic molecules that are catalyzed by other molecules but do not in turn catalyze other molecules. Because such parasitic molecules are in a majority, the reproduction of mutually catalytic molecules cannot be sustained.

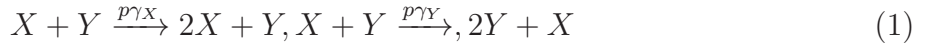
One possible way to resolve the problem of parasitism is to use compartmentalization[5–7]. When molecules are compartmentalized in a protocell, the unit of selection is shifted from a molecule to a protocell. Then, the protocells dominated by parasites are selected out, leaving behind protocells without parasites. However, such a compartmentalization introduces further complexity to the primitive form of life. It might be beneficial to use other molecules that act as a membrane; however, these molecules also need to be replicated. A specific reaction-diffusion mechanism to create compartments is also proposed[8, 9]. To avoid the further introduction of complexity, it is important to develop a simple mechanism for compartmentalization of a protocell.

Another important issue associated with protocells is the origin of heredity, i.e., the existence of specific molecules (such as DNA in the present cell) that are well preserved in the offspring cells and also control the behavior of the cells. By considering a simple hypercycle with only two chemicals, we have previously shown that the chemical with a slower synthesis rate and (as a result) that in minority plays the role of a heredity-carrier molecule. In fact, the relevance of such minority molecules in the hypercycle for evolvability and for the elimination of parasites is demonstrated[10, 11]. Here again, reaction in a compartment and the division of such a protocell was assumed.

Here, we address the following questions. Can we explain the origin of a protocell with compartmentalization and its growth-division cycle, in terms of a reaction-diffusion process

with minority molecules? How does the division process of protocells maintain synchrony with the replication of such minority molecules (i.e., what is the relationship between cell division and replication of DNA)? We address these questions by simply introducing spatial diffusion of molecules and catalytic reactions. We use the physicists' golden rule to study one the most simplest systems, i.e., a system consisting of only two chemical species that mutually catalyze the replication of each other, with the Brownian motion of molecules. From the results of a stochastic simulation, we show that the difference between the synthesis speed of the two molecular species leads not only to the formation of compartments as a cluster of such molecules but also to their recursive division with a certain cell size, even though no complicated mechanisms are assumed. Here, the localization of molecules around a minority molecule leads to compartmentalization, whereas infrequent replication of such a minority molecule induces the division of a compartment. Therefore, the reproduction of a protocell, which is essentially a cluster of molecules, progresses in synchrony with the replication of a minority molecule. We also demonstrate that this reproduction process tenaciously resists parasites.

We consider two molecular species that mutually catalyze the replication of each other. The reaction processes are expressed as follows:



The first process represents mutual catalytic replication with rate p , where the synthesis fractions of X and Y are γ_X and γ_Y , respectively. The second process represents the natural decomposition of X and Y with rates a_X and a_Y , respectively.

Because it is necessary to ensure discreteness in a molecular number, we adopt stochastic simulations rather than reaction-diffusion equations. In fact, the importance of ensuring discreteness in the molecular number has been discussed[12, 13]. We also take into account crowded molecules, and we assume that both X and Y molecules have a spherical shape with the same diameter (σ). These molecules are confined to a box with dimensions $L_x \times L_y \times L_z$, with elastic boundaries. Further, these molecules have the same mass and diffusion constant (D). Their motions obey the overdamped Langevin equations, which are numerically integrated by the Ermak-McCammon algorithm[14], such that

$$\Delta x = -\frac{D\Delta t}{k_B T} \nabla U(r) + S(t)$$

where $S(t)$ is a Gaussian white noise such that

$$\langle S(t) \rangle = 0, \langle S(t)S(t') \rangle = 2D\Delta t\delta(t - t').$$

with temperature $k_B T = 1$, where k_B denotes the Boltzmann constant. In the simulation, we set $\Delta t = 0.00005$. The interaction potential $U(r)$ between molecules is a Hertzian potential, $\phi(r) = E|\sigma - r|^{5/2}\Theta(\sigma - r)$, where $E = 10000\epsilon/\sigma^{5/2}$ with an energy unit ϵ , and the characteristic length scale σ (i.e., the diameter) of molecules and Θ is a Heaviside step function[15]. We start our simulation with the initial condition in which a single molecule Y is surrounded by X molecules[16].

At each time step, if the distance between every pair of X and Y is shorter than σ , the replication reaction occurs with a probability $p' = p/\Delta t$. In other words, p denotes the rate of replicating molecules per unit time under a crowded condition, i.e., when a pair of X and Y overlaps. If the reaction occurs, a new molecule X or Y is added to the system, with probability γ_X or γ_Y , respectively. Hence, the reaction rate for the replication of X and Y for a single reaction pair is given by $r_X \equiv p\gamma_X$ and $r_Y \equiv p\gamma_Y$, respectively. The position of the new molecule is chosen as $(x, y, z) = (x_0 + f_x, y_0 + f_y, z_0 + f_z)$, where (x_0, y_0, z_0) is the center between the reaction pair and (f_x, f_y, f_z) , which is randomly chosen from $-\sigma < f_x, f_y, f_z < \sigma$, by also avoiding overlap with other molecules. The decomposition process is carried out to remove all $X(Y)$ molecules, with a probability $a'_X = a_X/\Delta t$ ($a'_Y = a_Y/\Delta t$) at each step. Here, we fix the parameters as $\sigma = 1$, $D = 1$, and $p = 10^3$ and study the behavior of the system against the changes in the independent parameters γ_Y , a_X , and a_Y , while γ_X is set as $\gamma_X = 1 - \gamma_Y \geq \gamma_Y$ without loss of generality.

First, we examine the formation of a cluster by considering a single Y molecule and suppressing its replication and decomposition (i.e., $\gamma_Y = a_Y = 0$). If a_X is large, none of the X molecules remain to exist, whereas a stationary cluster of X molecules is formed around the single Y molecule when a_X is sufficiently small. For it, we fix $a_X = 4$ in the following. Indeed, we have computed the density distribution function N_X of X as a function of distance r from the single Y . The tail of the function for a large r is fitted well by the isotropic steady-state solution of the diffusion equation $N_X \sim \exp(-r/r_0)/r$ with $r_0 = \sqrt{D/a_X}$. For a small r , the value of the function is suppressed without $1/r$ -divergence owing to crowded molecules. Under the crowded condition, X and Y continuously overlap each other in the replication reaction. Then, the replication rate of X by a single Y molecule, denoted as R_X , is given

by $R_X = r_X \bar{n}_X$, where \bar{n}_X denotes the average number of X molecules that overlap with the single Y molecule. In the present simulations, $\bar{n}_X \sim 2$ [17]. Under the present crowded condition, the number of X molecules, N_{CX} , in a steady cluster is estimated by the balance between the replication and decomposition of X molecules, as $R_X = a_X N_{CX}$. By using $\bar{n}_X \sim 2$, we then estimate N_{CX} as ~ 500 , which is consistent with numerical results.

Now, we consider the replication of Y molecules with a rate that is slower than that of X . First, in Fig. 1, we show an example of the growth-division process of a cluster and provide the condition for it later. This process occurs as follows. For most of the time, the cluster maintains itself by replicating the X molecules, whereas the replication of the Y molecule rarely occurs. Once the Y molecule replicates, the distance between the two Y molecules is increased by diffusion, and these molecules contribute to the replication of X molecules. As the two Y molecules gradually move away from each other, the spatial distribution of X , synthesized by each Y , starts to elongate in the direction of the two Y molecules. Since the decomposition of the X molecule is dominant because it is located away from a Y molecule, the cluster of X molecules forms a dumbbell shape, and it finally splits into two (see also supplementary movies in [18]).

In fact, for this growth-division cycle to repeat, there is a restriction on the values of several parameters. First, the replication of Y should be sufficiently slow. After the replication of a new Y molecule, it takes some time for the two Y molecules to be separated at a distance of the size of the cluster. If another replication of Y occurs during this time, the cluster of X molecules cannot split into two because the replication of X molecules by the third Y molecule already begins. Second, the decay rate of Y should be sufficiently low. If this is not the case, the Y molecules would completely disappear before the division of a cluster. On the other hand, it should be noted that the cluster splits by the decomposition of X so that its rate a_X is higher than a_Y .

By taking several parameter values for γ_Y and a_Y , we have plotted the time evolution of the total number of molecules by starting from a single- Y initial condition in Fig. 2. We have found the following three typical behaviors: extinction, division, and explosion. In the extinction phase, all the molecules decay, and finally no molecules remain. In the parameters shown in this figure, extinction is initiated by the decomposition of a single Y molecule, followed by the extinction of X molecules because there are no more replications. Here, first, the number of molecules fluctuates at around a constant value, corresponding to

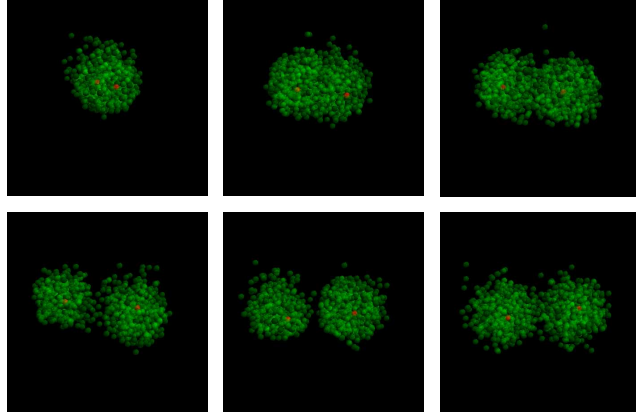


FIG. 1: Snapshots after the replication of Y . The green semitransparent particles represent the X molecules. The red particles represent the Y molecules, located deep within the clusters. Time evolves from the left top to right bottom, as $t = 0.1, 0.5, 1, 1.5, 2, 2.5$. The parameter values are $r_Y = 0.01$, $a_X = 4$, and $a_Y = 0.002$.

a single cluster, and then, it suddenly decreases with the disappearance of the Y molecule. (When a_X is considerably larger than a_Y , the extinction of X molecules precedes that of Y .)

In the division behavior, a stepwise increase in the total number of molecules is observed. For reference, we also plot dotted lines in Fig.2 at the level of several multiples of the number of molecules in a single cluster. The total number of molecules fluctuates around one of the dotted lines and gradually increases, as is consistent with the division process of a certain size of a cluster. In the explosion case, the number of molecules show an exponential increase without a step-like behavior. The number of molecules increases by maintaining a single cluster. Typical snapshots of the division and explosion cases are shown in Fig. 3, where the green particles represent the X molecules.

In Fig. 3, a phase diagram of these three behaviors is plotted on the r_Y - a_Y plane by fixing the decay rate of X at $a_X = 4$. The three phases are separated in the parameter space. The division phase lies at smaller r_Y and a_Y . An increase in a_Y leads to extinction, whereas an increase in r_Y leads to explosion. Near the boundary between the division and the extinction phases, the corresponding two behaviors coexist, depending on each sample.

The boundaries between the phases are estimated as follows. In the case of the boundary of the extinction phase, we note the balance between the replication and the decay of Y .

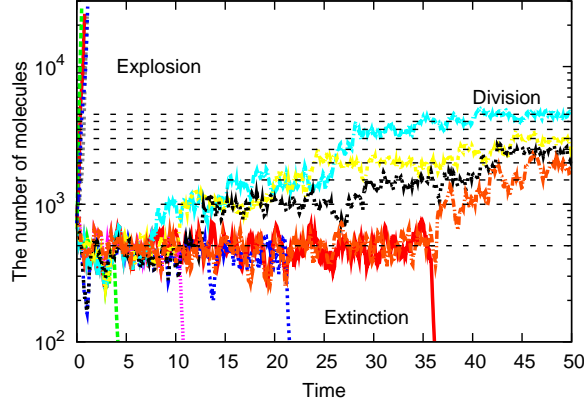


FIG. 2: Time evolution of the total number of molecules. Data are obtained from the following three sets of parameters with several samples; $r_Y = 0.001, a_Y = 0.2$ (leading to extinction), $r_Y = 0.01, a_Y = 0.002$ (division), and $r_Y = 1, a_Y = 0.02$ (extinction). Here, we set $a_X = 4$. The dotted lines represent several multiples of the molecule number of a single cluster, N_{CX} .

The replication rate of a Y molecule in the cluster is estimated by $\sim R_Y = r_Y \bar{n}_X$, and the decay rate is estimated by $\sim a_Y$. In the case of the division phase, the replication rate of Y should be greater than its decay rate so that the region satisfies

$$a_Y < R_Y. \quad (3)$$

This boundary is plotted in the r_Y - a_Y plane, which is consistent with our simulation results.

The boundary between the division and the explosion phases is estimated by the balance between the replication rate of Y and the size of the cluster. The linear size of a single cluster, L_C , is estimated by $L_C \sim N_{CX}^{1/3}$. The division process starts by the replication of Y and is completed when the two Y molecules diffuse approximately to the distance L_C . We denote the typical timescale of dividing processes by τ_D , and we obtain $L_C^2 \sim D_Y \tau_D$, where D_Y denotes the effective diffusion constant of Y . Here, D_Y is slightly increased from $D = 1$ because of the repulsive interactions with X molecules, and it is estimated as $D_Y \sim 3$ from the simulations for Fig.3..

At the phase boundary, only a single replication of Y should occur during this time τ_D to complete the division, so that $R_Y \tau_D = 1$. Here, $\tau_D = L_C^2 / D_Y = (R_X / a_X)^{2/3} / D_Y$. Thus, the boundary is expressed as

$$R_Y = D_Y \left(\frac{a_X}{R_X} \right)^{2/3}. \quad (4)$$

The estimated boundary is plotted in Fig. 3, which is also consistent with our results.

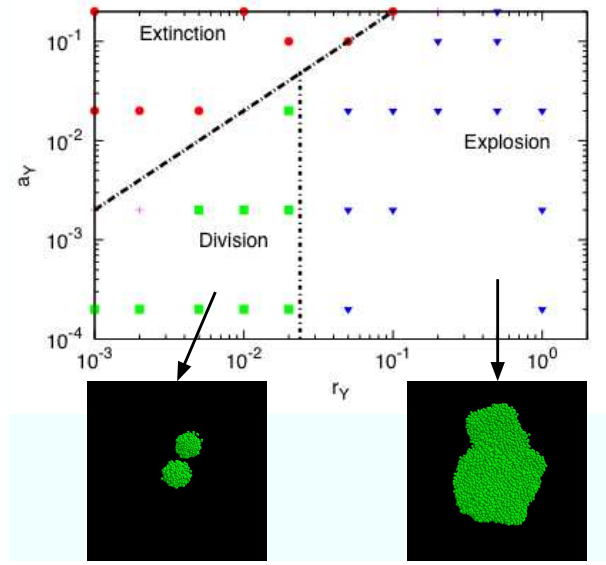


FIG. 3: Phase diagram of our model in r_Y - a_Y space. The parameters are $p = 10^3$ and $a_X = 4$. The plane is divided into the following three regions: extinction (\circ), division (\square), and explosion (∇). At the parameters with $+$ points, two behaviors of the adjacent regions coexist. We also plot the theoretically estimated boundaries given by eqs. (3) and (4).

In summary, we have studied a simple reaction system of two mutually catalytic molecules that diffuse under a crowded condition. When the replication speed considerably differs between the two molecular species, the molecule with a slower replication rate becomes a minority molecule, and the minority molecule leads to the formation of a cluster of molecules and its division into two clusters as a result of replication of the molecule. This division is repeated to produce protocells of a certain size. Here, we have presented the simulation results for a three-dimensional case; however, we have confirmed the existence of the present three phases, in particular, the division of a protocell in a two-dimensional case.

To further examine whether the present protocell resists parasitic molecules, we have also extended our model to include parasitic molecules X' and Y' , which are catalyzed by other molecular species but cannot catalyze the replication of other molecules. If the variety and/or replication rates of the parasites are large, they would dominate. Protocells dominated by parasites, however, stop growing and eventually disappear, whereas cells that are not infected by parasites continue to grow and divide (see Supplementary Material and Movie[18]). In particular, it is observed that when parasite Y' appears, it is spontaneously emitted from the cluster, so that Y' parasites are eliminated. This is because Y' cannot

synthesize X ; therefore, it cannot remain at the center of the cluster, and it diffuses out of the cluster.

It should be noted that the reaction scheme given in eq. (1) is extremely simplified. For such reactions to progress, some resources are necessary for the synthesis of X and Y . If such resources are sufficiently supplied, the original model (eq. (1)) is derived. In reality, however, these resources have to be supplied from the outside[19]. As the size of the cluster increases, their supply cannot be penetrated deeper into the cluster; therefore, replication is limited only at the periphery, whereas decomposition occurs in a bulk. Hence, the growth of a cluster without division stops when the cluster achieves a certain size. In contrast, when the cluster divides into two before it reaches the size threshold, the molecules continue to replicate.

In conclusion, the formation of a compartment and its growth-division cycle are shown to be a natural outcome of mutually catalytic reactions with a minority molecule species. In addition, this division of a protocell is synchronized with the replication of the minority molecule. As already discussed, the minority molecule Y can act as a carrier of heredity, as it is preserved well, controls the characteristic of the protocells, and gives evolvability[10, 11, 20]. Here, we have shown that the replication of this minority molecule is synchronized with the division of a protocell. These characteristics of the minority molecule Y agree well with those required by genetic information, played by DNA in the present cell. The question about the origin of genetic information from catalytic reactions, addressed by Dyson[2], is thus answered.

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Supplementary Material for
Reproduction of a Protocell by Replication of Minority Molecule
in Catalytic Reaction Network

In this Supplementary, we show that the protocell at the division phase in our model offers resistance to parasitic molecules, which are synthesized with the help of other molecules, but cannot catalyze other molecules. Molecules X and Y are usually polymers, with a sequence of monomers. Generally, there are errors in the replication of a (polymer) molecule; therefore, the sequence in X or Y can be modified. After the modification of the sequence, the molecules most likely lose their catalytic activity, since such functional molecules are rather rare. These molecules are often synthesized by other molecules, and thus, they are known as parasites. Hence, the parasitic molecule X' (Y') replaces the original molecule with a certain rate; this replacement process is regarded as the mutation of X (Y). In the replication processes of X (Y), parasitic molecules such as X' (Y'), and not X (Y), are synthesized with a probability μ . Because the sequence that has a catalytic activity for the synthesis of Y (X) is rare, most mutants are parasites; therefore, backward mutations from X' (Y') to X (Y) are neglected.

After the appearance of parasitic molecules, they replicate in the same way as the original X and Y molecules do.



In this supplementary, we assume that the replication and decay rates of parasitic molecules are identical with the original molecules; however, it is observed that several simulations with different values give similar behaviors. Time evolution is simulated from the initial condition, where a single Y molecule is located at the center of a few dozens of X molecules. When the mutation probability μ is considerably high, the parasitic molecules dominate and they stop growing in some time. When μ is not so high, the compartment splits into two before the parasites accumulate completely, so that the compartment with a non-parasitic molecule pair continues to grow.

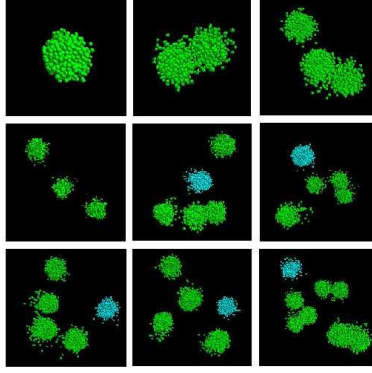


FIG. 4: Time evolution of the system with parasitic molecules. The green and light blue particles represent the X and X' molecules, respectively. Snapshot patterns at $t = 5, 10, 15, 20, 30, 35, 40, 45, 50$ are plotted from the left top to right, left middle to right middle, and left bottom to right bottom. Compartments with X molecules continue to grow whereas those covered by X' do not grow. Parameters are $p = q = 10^3$, $r_Y = 0.02$, $a_X = 4$, $a_Y = 2 \times 10^{-3}$, $\mu = 0.0001$.

An example of temporal evolution is shown in Fig. 4. The number of molecules increase in time, and the cluster divides as in the case of the original model. At some time (corresponding to the center middle box), parasitic molecules X' appear by mutations and they replace the host molecule X . Even if a compartment is dominated by parasites X' , other compartments with non-parasitic pairs can continue to grow and divide.

We have also plotted the time evolution of the number of non-parasitic and parasitic molecules for several samples in Figs. 5 and 6, respectively. In Fig. 5, from the initial condition, the number of X molecules fluctuates around the number of single clusters, and some samples show a stepwise increase, as observed in our original model. However, a sudden decrease in the number of X molecules is also observed, and this decrease corresponds to the appearance of parasitic molecules in Fig. 6. The number in Fig. 6 also shows stepwise increases corresponding to the number of a single cluster of X' . These stepwise increases in X' are caused by the transfer of non-parasitic pairs from some other cluster, and are not the autonomous growth by the replications of Y within the cluster of the pair of X' and Y .

On the other hand, an example of resistance to the parasite Y' is shown in Fig. 7. A movie is also available in [1]. The Y molecule is centered around a cluster of X molecules since it helps the synthesis of X molecules. In contrast, parasitic Y' molecules cannot aid the synthesis of X ; therefore, X molecules do not gather in the vicinity of Y' and Y' freely

diffuses until it goes out of the cluster (see Fig. 7). Once the parasite Y' diffuses out of the cluster, it cannot replicate without the help of X , and therefore, it decomposes and eventually disappears. Although we have assumed only mutual catalysis and diffusion of molecules, parasitic molecules are selectively removed out of a cluster.

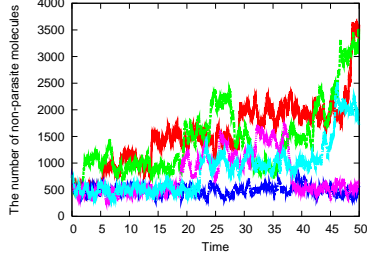


FIG. 5: Time evolution of the number of non-parasitic molecules. Data are obtained for several samples, as plotted in different colors. The red line represents the sample shown in Fig. 4.

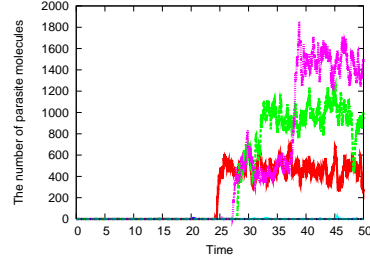


FIG. 6: Time evolution of the number of parasitic molecules for the same set of samples shown in Fig. 5, plotted with corresponding colors.

[1] <http://bopper.t.u-tokyo.ac.jp/~kamimura/movie.html>

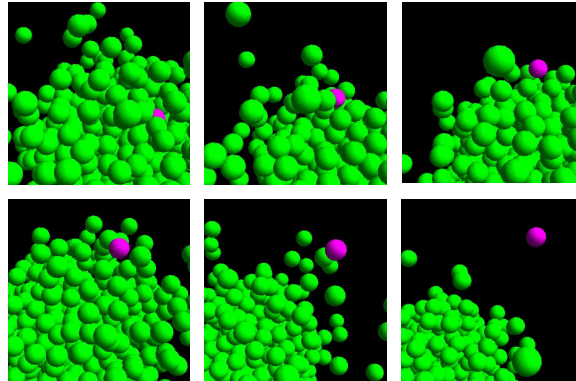


FIG. 7: Time evolution of the system with parasitic molecules Y' . The green and magenta particles represent the X and Y' molecules, respectively. Snapshot patterns at $t = 1, 1.5, 2, 2.5, 3, 3.5$ are plotted from the left top to right and left bottom to right bottom.